

This is a “preproof” accepted article for Weed Science. This version may be subject to change in the production process, *and does not include access to supplementary material*.

DOI: 10.1017/wet.2025.10053

Pervasive weed management and soybean yield with 2,4-D and glufosinate applied alone, mixed or sequentially

Eric A. L. Jones¹, Jill K. Alms², David A. Vos²

¹ Assistant Professor, Agronomy, Horticulture, and Plant Science Department, South Dakota State University, Brookings, South Dakota, USA

² Agricultural Research Manager, Agronomy, Horticulture, and Plant Science Department, South Dakota State University, Brookings, South Dakota, USA

Corresponding author: Eric Jones; E-mail: eric.jones@sdstate.edu

This is an Open Access article, distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives licence (<http://creativecommons.org/licenses/by-nc-nd/4.0/>), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is unaltered and is properly cited. The written permission of Cambridge University Press must be obtained for commercial re-use or in order to create a derivative work.

Abstract

Field experiments were conducted near Beresford and South Shore, South Dakota, in 2023 and 2024 to determine weed control and soybean yield with 2,4-D and glufosinate applied alone, mixed, and sequentially. Sequential applications were made 12 d after the initial application. 2,4-D plus glufosinate additively controlled and reduced the height of all tested weed species. Sequential applications increased common lambsquarters, waterhemp, redroot pigweed, and velvetleaf control compared with 2,4-D, glufosinate, or 2,4-D plus glufosinate applied alone. The herbicide order in the sequential application did not influence broadleaf weed control. Yellow foxtail control was greater with sequential applications of glufosinate. Soybean yield at Beresford was similar across all treatments. Yields were generally greater with sequential herbicide applications, where glufosinate was applied initially at South Shore. The experiment results suggest that weed control and soybean yield are greater with 2,4-D plus glufosinate or sequential application treatments utilizing 2,4-D and glufosinate.

Nomenclature: 2,4-D; glufosinate; common lambsquarters, *Chenopodium album* L.; common waterhemp, *Amaranthus tuberculatus* Moq. J.D. Sauer; redroot pigweed; *Amaranthus retroflexus* L.; velvetleaf, *Abutilon theophrasti* L.; yellow foxtail, *Setaria pumila* (Poir.) Roem. & Shult.

Keywords: herbicide interactions; herbicide resistance; weed management; sequential applications

Introduction

Herbicides are recommended for application in mixtures to increase efficacy, application efficiency, control spectrum, and to reduce selection pressure on herbicide-resistant weed biotypes (Green 1989; Renton et al. 2024). Various herbicide mixtures can provide activity that is antagonistic, additive, or synergistic on select weeds (Colby 1967; Green 1989). The resulting activity can be an effect of chemical incompatibility or physiology of the herbicides (Barbieri et al. 2022; Green 1989). Physiological antagonism or synergism can occur when herbicides with different modes of action are mixed and counteract *in planta* negatively or positively (Meyer et al. 2019; Ou et al. 2018). Since mixtures and sequential applications of various herbicides are often recommended, understanding how the mixtures or sequential applications of these herbicides perform on various weed species under field conditions is critical.

2,4-Dichlorophenoxyacetic acid (WSSA Group 4) and glufosinate (WSSA Group 10) applied alone, mixed, and sequentially may become more common for weed management due to herbicide resistance and the commercial availability of tolerant soybean varieties (Shyam et al. 2021). 2,4-Dichlorophenoxyacetic acid is a slow-acting, phloem-mobile herbicide with activity predominantly on broadleaf weeds and kills plants by derepressing transcription factors from auxin receptors to increase auxin concentrations to cause growth malformations, increase excessive production of reactive oxygen species and induction of stress responses to divert away from essential processes/constituents, which lead to plant death due to chloroplast destruction (Grossman 2010; Peterson et al. 2016). Glufosinate is a fast-acting, contact herbicide with activity on some broadleaf and grass weeds (Corbett et al. 2003). Glufosinate inhibits glutamine synthetase (EC 6.3.1.2), which in turn inhibits the production of photosynthesis precursors, leading to the production of reactive oxygen species that ultimately disrupt cell membrane integrity (Takano et al. 2020). The mixture of 2,4-D and glufosinate is a labeled application in soybean (Anonymous 2023; Anonymous 2024). Since 2,4-D and glufosinate have different modes of action, research providing information regarding whether this mixture is antagonistic, additive, or synergistic on various weed species is critical for effective weed management. Sequential applications of these herbicides may be utilized in soybean as well. 2,4-dichlorophenoxyacetic acid and glufosinate have label restrictions limiting sequential applications to occur 12 and 5 d after the initial herbicide application, respectively (Anonymous

2023; Anonymous 2024). Investigations that assess the interaction of sequential applications of 2,4-D and glufosinate should include application timings reflecting the most restrictive labeled application time.

Waterhemp is the most troublesome weed in the Midwest United States due to the biology of the plant and widespread evolution of herbicide resistance (Butts et al. 2018; Jones et al. 2019). Waterhemp has evolved resistance to herbicides from Group 2, 4, 5, 6, 9, 14, 15, and 27 and multiple herbicide-resistant populations are common (Faleco et al. 2022; Heap 2025). 2,4-Dichlorophenoxyacetic acid resistance has been confirmed in Illinois, Minnesota, Missouri, and Nebraska (Bernards et al. 2012; Evans et al. 2019; Shergill et al. 2018; Singh et al. 2024). While glufosinate resistance is not yet confirmed in waterhemp, control failures have been reported (Hamberg et al. 2023; Landau et al. 2025). 2,4-dichlorophenoxyacetic acid and glufosinate applied alone, mixed, and sequentially have been previously studied on waterhemp. The research determined that 2,4-D plus glufosinate provided additive control, and sequential applications were more efficacious than single herbicide applications (Craigmyle et al. 2013a; Craigmyle et al. 2013b; Haarman et al. 2020).

While waterhemp is the focus of many weed management plans, other weed species are usually present in sufficient population densities to require management. Therefore, it is of interest to determine the effectiveness of various 2,4-D and glufosinate treatments on other common weed species. 2,4-Dichlorophenoxyacetic acid has no activity on grass weed species and may increase grass weed population densities if applied extensively (Hodgskiss et al. 2022). Currently, there are no published data on the effectiveness of 2,4-D and glufosinate mixed or applied sequentially on common lambsquarters, redroot pigweed, yellow foxtail, and velvetleaf which are historically pervasive soybean weeds (Shurtlee and Coble 1985; Staniforth 1965; Stoller and Woolley 1985). The objectives of this research were to determine if 1) 2,4-D plus glufosinate were additive, antagonistic, or synergistic on these weeds, 2) compare the effectiveness of sequential applications with singular herbicide applications, and 3) the herbicide treatments impacted soybean yield.

Materials and Methods

Field experiments were conducted in 2023 and 2024 at Beresford (43.050067, -96.896512) and South Shore (45.106553, -97.095680), SD, for a total of four site-years. The soil at the Beresford location is an Egan-Trent silty clay loam (Fine-silty, mixed, superactive, mesic Pachic/Udic Haplustoll) and a Kranzburg-Brookings silty clay loam (Fine-silty, mixed, superactive, frigid Calcic/Pachic Hapludoll) at the South Shore location. Common lambsquarters, waterhemp, and velvetleaf populations occurred at Beresford (two site-years) and populations of redroot pigweed and yellow foxtail occurred at South Shore (two site-years). Each site was tilled with a field cultivator prior to experiment establishment. All fields were under a corn-soybean rotation, Soybean seeds were planted at population density of 395,000 seeds ha⁻¹ with 76 cm row spacing for all experiments. The soybean varieties DSR-150SE (Dairyland Seed Company, Corteva Agriscience, Indianapolis, IN) and NK09-H7E3 (NK Seeds, Syngenta, Greensboro, NC) were planted on May 17th and May 23rd at Beresford and South Shore in 2023, respectively. Soybean varieties AE1900 (Mustang Seeds, M.S. Technologies, West Point, IA) and AE1030 (Mustang Seeds, M.S. Technologies, West Point, IA) were planted on May 16th and June 7th at Beresford and South Shore in 2024, respectively. Weather data for each site year is provided in Table 1. Preemergence herbicides were omitted to ensure the maximum weed emergence.

Treatments were arranged in a randomized complete block design with four replications. Individual plots were 3 m wide × 12 m long. Herbicide treatments are provided in Table 2. Herbicide treatments were applied to plots with a CO₂-pressurized backpack sprayer calibrated to deliver 140 L ha⁻¹ at 165 kPa while traveling at 4.8 km hr⁻¹ and 46 cm above the target weed height. Weeds were approximately 15 cm (7.6 to 56 cm in 2023; 5 to 38 cm in 2024) in height at the time of treatment. The 15 cm height target was selected based on the 2,4-D label despite the glufosinate label recommending treating weeds at a target height of 7.6 cm (Anonymous 2023; Anonymous 2024). The sequential herbicide applications were made 12 days after the initial herbicide application (DAIT). The weeds were approximately 50 cm in height at the sequential application (20 to 91 cm in 2023; 35 to 66 cm in 2024). All treatments were applied with TeeJet 8003 AIXR spray nozzles (TeeJet® Technologies, Spraying Systems, Wheaton, IL, USA). These nozzles were selected for consistency across the herbicide treatments based on the 2,4-D label

(Anonymous 2024). 2,4-Dichlorophenoxyacetic acid choline (Enlist One, Corteva AgriscienceTM, Indianapolis, IN, USA) was applied at 1165 g ae ha⁻¹ for all 2,4-D treatments, while glufosinate (Liberty, BASF, Raleigh, NC, USA) was applied at 655 g ai ha⁻¹ with 10 g L⁻¹ ammonium sulfate for all glufosinate treatments. Clethodim (560 g ai ha⁻¹) was applied with the spray parameters as described above to 2,4-D-only treatments 21 DAIT to control grass weeds that could confound the control for broadleaf weeds. No response variable data were recorded for grass species in these plots.

Weed control evaluations were made using visual estimates based on a scale ranging from 0% to 100%, where 0% equals no control (i.e., no injury symptoms on any tissue) and 100% equals complete control (i.e., total necrosis). Weed height reduction was determined by measuring from the soil surface to the apical growing point for three representative plants of each species arbitrarily selected in the central region of each plot. Percentage height reduction was calculated by dividing the heights of the plants in the treated plots by the heights of the plants in the nontreated plots. Control and height reduction evaluations were made 28 DAIT. Soybeans were harvested after reaching physiological maturity using a combine and the yield was adjusted to 13% moisture.

Control, height reduction and soybean yield data were subjected to analysis of variance (ANOVA) using the Glimmix procedure in SAS 9.4 (Statistical Analysis Software Institute, Cary, NC, USA) ($\alpha = 0.05$). Herbicide treatment was considered a fixed effect, whereas block and year and their interactions were considered random effects. Year was considered random to allow inferences to be made across broader conditions and locations (Blouin et al. 2011; Moore and Dixon 2015). Treatment means were separated using Fisher's Least Significant Difference test ($P \leq 0.05$).

2,4-Dichlorophenoxyacetic acid plus glufosinate mixtures were evaluated to determine if the resultant activity was additive, antagonistic, or synergistic 28 DAIT using Colby's Method (Colby, 1967). Colby's method calculates an expected control value for an herbicide mixture based on the control of the individual herbicides and the expected control value is compared with the control of the tested mixture. 2,4-D plus glufosinate treatments were analyzed using the equation for Colby's method [1]:

$$E = (XplusY) - (\frac{xy}{100}) [1]$$

where E is expected % control of two herbicides applied in a mixture, X is % control of X herbicide when applied alone, and Y is % control of Y herbicide when applied alone. The expected control was compared with the observed control using a two-sided t-test ($\alpha = 0.05$). If the control was greater than the expected % control, the mixture was considered to be synergistic, whereas if the % control was lower than the expected % control, the mixture was antagonistic (Colby 1967). If the observed and expected % controls were equal, the mixture was considered additive (Colby 1967). Since 2,4-D does not control yellow foxtail, statistical deviations from the single and mixed treatments of 2,4-D and glufosinate can provide evidence of either antagonism or synergism (Flint and Barrett 1989; Meyer and Norsworthy 2019). Height reduction data of the broadleaf weeds were subjected to Colby's Method as well. The control of sequential herbicide treatments was compared to the control of the 2,4-D plus glufosinate mixture to determine if activity was antagonistic or synergistic (Burke et al. 2005).

Results and Discussion

Velvetleaf

2,4-D and N fb 2,4-D provided the least control when compared with the other herbicide treatments (Table 3). However, no treatment provided less than 90% velvetleaf control suggesting all herbicide treatments were effective. The sequential herbicide applications reduced velvetleaf height more than the single by 20% or more (Table 4). 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate reduced velvetleaf height similar to some sequential herbicide applications (Table 4). 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate were determined to provide additive control and height reduction (Table 5).

Redroot pigweed

Glufosinate and 2,4-D plus glufosinate controlled redroot pigweed approximately 10% more than 2,4-D (Table 3). Similarly, N fb glufosinate and N fb 2,4-D plus glufosinate provided 30% more control than N fb 2,4-D (Table 3). All sequential herbicide applications provided more redroot pigweed control than single herbicide applications except for 2,4-D fb 2,4-D where control was approximately 8% less than single herbicide (Table 3).

2,4-D and 2,4-D plus glufosinate reduced redroot pigweed height approximately 15% more than glufosinate (Table 4). All N fb other herbicide treatments reduced redroot pigweed height similarly (Table 4). Height reduction with sequential herbicide applications followed a similar trend when compared with control (Tables 3 and 4). The treatments 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate were determined to be additive for control and height reduction of redroot pigweed (Table 5).

Waterhemp

2,4-D and 2,4-D plus glufosinate provided more waterhemp control than glufosinate and control was less for N fb 2,4-D compared with 2,4-D (Table 3). Glufosinate was not as effective at controlling waterhemp as N fb glufosinate and 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate provided similar waterhemp control (Table 3). Most sequential herbicide applications improved common waterhemp control by 7 to 30% compared with a single herbicide (Table 3).

Waterhemp height reductions followed a similar pattern compared with control (Table 4). Waterhemp control with 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate was determined to be additive (Table 4). The effect of 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate on waterhemp height was determined to be additive (Table 5).

Common lambsquarters

2,4-D and 2,4-D plus glufosinate provided 12 to 16% more control of common lambsquarters than glufosinate (Table 3). The no initial herbicide (N) followed by (fb) 2,4-D plus glufosinate improved common lambsquarters control by approximately 8% more than N fb 2,4-D and N fb glufosinate (Table 3). Sequential herbicide applications provided similar common lambsquarters control to 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate. However, the sequential applications provided 12% greater common lambsquarters control compared with glufosinate, N fb 2,4-D, and N fb glufosinate (Table 3).

Glufosinate and N fb 2,4-D plus glufosinate resulted in the least common lambsquarters height reduction (Table 4). 2,4-D, 2,4-D plus glufosinate, N fb 2,4-D and N fb glufosinate reduced common lambsquarters height similarly. Sequential applications reduced height the greatest (Table 3). Common lambsquarters control and height reduction was determined to be additive for 2,4-D plus glufosinate and N fb 2,4-D plus glufosinate (Table 5).

Yellow foxtail

Two glufosinate applications provided greater yellow foxtail control than one glufosinate application (Table 3). When glufosinate was applied once, control was greater with the 2,4-D fb glufosinate (79%) compared with glufosinate fb 2,4-D (63%). Similarly, yellow foxtail height reduction with two applications of glufosinate was approximately 26% greater than treatments with only one application of glufosinate (Table 4). 2,4-D plus glufosinate and glufosinate controlled and reduced the height of yellow foxtail similarly which suggests that the tank mixture has additive activity (Tables 3 and 4). The same trend was seen with the N fb 2,4-D plus glufosinate and N fb glufosinate treatments further suggesting additive activity (Tables 3 and 4).

Soybean Yield

Due to a significant interaction between location and treatment ($P = 0.008$) and differences in the weed species composition at each location, soybean yield data were analyzed by location. Only two herbicide treatments yielded differently at Beresford; 2,4-D plus glufosinate fb 2,4-D and N fb 2,4-D (Table 6). All other treatments yielded similarly and higher than the non-treated control (Table 6).

Yield was higher at South Shore with two sequential applications of glufosinate (Table 6). These results of higher yield with two sequential applications of glufosinate correspond with increased yellow foxtail control (Tables 4, 5, and 6). Treatments with one application of glufosinate yielded similarly (Table 6). The yield from 2,4-D-only treatments at South Shore are likely not true representations of yield as clethodim was applied to control yellow foxtail. Therefore, yield would likely be lower if only 2,4-D was applied to a field with yellow foxtail and/or other grass weed species.

Discussion

Control on waterhemp in the presented research was comparable with previous research utilizing similar treatments (Craigmyle et al. 2013; Duenk et al. 2023). The waterhemp control estimates for the glufosinate and N fb glufosinate were counterintuitive as the N fb glufosinate provided greater control; glufosinate effectiveness decreases as weed size increases (Steckel et al. 1997). The separation of control by these two treatments is likely a function of plant growth

after treatment. Waterhemp plants treated with glufosinate had approximately two weeks longer to exhibit regrowth compared with plants treated with N fb glufosinate (Haarmann et al. 2020; Jones et al. 2024). Despite the control separation, neither of these treatments was effective on waterhemp, further bolstering the label to treat plants at 7.6 cm in height and using nozzles that create fine droplets to increase coverage (Anonymous 2023).

While common lambsquarters, redroot pigweed, and velvetleaf control have not previously been reported with mixtures or sequential applications of 2,4-D and glufosinate, the control with single herbicide applications from previous research is similar to the results presented (Coetzer et al. 2002; Fawcett and Slife 1978; Robinson et al. 2012; Steckel et al. 1997). Yellow foxtail control from this research was much less compared with previous research utilizing similar glufosinate rates and weed size (Corbett et al. 2003; Hamill et al. 2000). Additive control and height reduction was determined in the presented research for all tested weed species, which is concordant with previous research on similar weed species (Craigmyle et al. 2013b; Merchant et al. 2013; Meyer and Norsworthy 2019).

Sequential herbicide applications increased the control and height reductions of the broadleaf weed species compared to 2,4-D and glufosinate applied alone, but the order in which the herbicides were applied did not influence efficacy. The order of herbicides in sequential applications was important for yellow foxtail control and height reduction. Control and height reduction of yellow foxtail was greater when glufosinate was applied in both applications, which is similar to previous research (Jones et al. 2022). Common lambsquarters, redroot pigweed, and velvetleaf control when treated with 2,4-D plus glufosinate was like the sequential applications. The height reduction of these species followed a similar trend, excluding redroot pigweed, where height reduction was greater with sequential applications. Waterhemp and yellow foxtail control and height reduction were greater with sequential herbicide applications compared with 2,4-D plus glufosinate. Therefore, recommendations for applying 2,4-D plus glufosinate or these herbicides sequentially should be made based on the weed species present in the specific field. This conclusion is further reinforced by reduced soybean yields with herbicide treatments at South Shore that provided poor yellow foxtail control.

Practical implications

2,4-D and glufosinate applied sequentially or mixed were more effective in managing the weed community as a whole at the locations of the experiments. Since the herbicide application order did not influence the effectiveness for sequential applications to manage broadleaf species, both 2,4-D and glufosinate should be utilized to reduce selection pressure on resistant biotypes. The mixture of 2,4-D plus glufosinate is likely the most utilitarian, as sequential applications rely on ideal conditions between the applications. This recommendation is further bolstered since soybean yields were generally higher when the mixture was applied or when both herbicides were used in a sequential application treatment. While yellow foxtail management was more effective with two glufosinate applications, this species should be managed with other effective herbicides or nonchemical tactics. These results further reinforce that weeds should be treated at a smaller size (<10 cm) with these herbicides as the N fb herbicide treatments were less effective on the tested weed species and resulted in low soybean yield. Recommendations for these herbicides should focus on timely application (i.e., 7.6 vs >15 cm weeds), using the most appropriate nozzle (i.e., fine vs coarse droplet) and output for coverage (i.e., 140 vs 187 L ha⁻¹).

Acknowledgements

The authors extend thanks to Allen Heuer (Northeast Research Farm), Bradley Rops (Southeast Research Farm), and Josalyn Fousert (Southeast Research Farm) for technical support. The authors also thank Dr. Micheal D.K. Owen for reviewing the manuscript prior to submission.

Funding

Project funding was provided by the South Dakota Soybean Research and Promotion Council.

Competing interests

The authors declare no competing interests.

References

- Barbieri GF, Young BG, Dayan FE, Streibig JC, Takano H, Merotto A jr, Avila LA (2022) Herbicide mixtures: interactions and modeling. *Advances in Weed Science* 40:e020220051
- Bernards ML, Crespo RJ, Kruger GR, Gaussoin R, Tranel PJ (2012) A waterhemp (*Amaranthus tuberculatus*) population resistant to 2,4-D. *Weed Sci* 60:379–384
- Butts TR, Vieira BC, Latorre DO, Werle R, Kruger GR (2018d) Competitiveness of herbicide-resistant waterhemp (*Amaranthus tuberculatus*) with soybean. *Weed Sci* 66:729–738
- Coetzer E, Al-Khatib K, Peterson DE (2002) Glufosinate efficacy on *Amaranthus* species in glufosinate-resistant soybean (*Glycine max*). *Weed Technol* 16:326–331
- Colby SR (1967) Calculating synergistic and antagonistic response of herbicide combinations. *Weeds* 15:20–22
- Corbett JL, Askew SD, Thomas WE, Wilcut JW (2004) Weed efficacy evaluations for bromoxynil, glufosinate, glyphosate, pyriithiobac, and sulfosate. *Weed Technol* 18:443–453
- Craigmyle BD, Ellis JM, Bradley KW (2013a) Influence of herbicide program on weed management in soybean with resistance to glufosinate and 2,4-D. *Weed Technol* 27:78–84
- Craigmyle BD, Ellis JM, Bradley KW (2013b) Influence of weed height and glufosinate plus 2,4-D combinations on weed control in soybean with resistance to 2,4-D. *Weed technol* 27:271–280
- Duenk E, Soltani N, Miller RT, Hooker DC, Robinson DE, Sikkema PH (2023) Multiple-herbicide-resistant waterhemp control in glyphosate/glufosinate/2,4-D-resistant soybean with one- and two-pass weed control programs. *Weed Technol* 37:34–39
- Evans CM, Strom SA, Riechers DE, Davis AS, Tranel PJ, Hager AG (2019) Characterization of a waterhemp (*Amaranthus tuberculatus*) population from Illinois resistant to herbicides from five sites-of-action. *Weed Technol* 33:400–410

- Faleco FA, Oliveira MC, Arneson NJ, Renz M, Stoltenberg DE, Werle R (2022) Multiple herbicide resistance in waterhemp (*Amaranthus tuberculatus*) accessions from Wisconsin. *Weed Technol* 36:597–608
- Fawcett RS, Slife FW (1978) Effects of 2,4-D and dalapon on weed seed production and dormancy. *Weed Sci* 26:543–547
- Green JM (1989) Herbicide antagonism at the whole plant level. *Weed Technol* 3:217–226
- Grossmann K (2010) Auxin herbicides: current status of mechanism and mode of action. *Pest Manag Sci* 66:113–120
- Haarman JA, Young BG, Johnson WG (2020) Control of waterhemp (*Amaranthus tuberculatus*) regrowth after failed applications of glufosinate or fomesafen. *Weed Technol* 34:794–800
- Hamberg RC, Yadav R, Owen MDK, Licht MA (2022) Differential susceptibility of Iowa waterhemp (*Amaranthus tuberculatus*) populations to 2,4-D, dicamba, and glufosinate. *Can J Plant Sci* 103:595–599
- Hamill AS, Knezevic SZ, Chandler K, Sikkema PH, Tardif FJ, Shrestha A, Swanton CJ (2000) Weed control in glufosinate-resistant corn (*Zea mays*). *Weed Technol* 14:578–585
- Hodgskiss CL, Legleiter TR, Young BG, Johnson WG (2022) Effects of herbicide management practices on the weed density and richness in 2,4-D-resistant cropping systems in Indiana. *Weed Technol* 36: 130–136
- Jones EAL, Bradshaw CL, Contreras DJ, Cahoon CW, Jennings KM, Leon RG, Everman WJ (2024a) Growth and fecundity of Palmer amaranth escaping glufosinate in soybean. *Weed Technol* 38:e43
- Jones EAL, Leon RG, Everman WJ (2022) Control of pervasive row crop weeds with dicamba and glufosinate applied alone, mixed, or sequentially. *Weed Technol* 36:733–739
- Jones EAL, Owen MDK, Leon RG (2019) Influence of multiple herbicide resistance on growth in *Amaranthus tuberculatus*. *Weed Res* 59:235–244
- Landau C, Bradley K, Burns E, DeWerff R, Dobbels A, Essman A, Flessner M, Gage K, Hager A, Jhala A, Johnson PO, Johnson W, Lancaster S, Lingenfelter D, Loux M, Miller E,

- Owen MDK, Sarangi D, Sikkema P, Sprague C, VanGessel M, Werle R, Young B, Williams M (2025) Weather and glufosinate efficacy; a retrospective analysis looking forward to the changing climate. *Weed Sci In press*
- Merchant RM, Sosnoskie LM, Culpepper AS, Steckel LE, York AC, Braxton LB, Ford JC (2013) Weed response to 2,4-D, 2,4-DB, and dicamba applied alone or with glufosinate. *J Cotton Sci* 17:212–218
- Meyer CJ, Norsworthy JK (2019) Influence of weed size on herbicide interactions for Enlist™ and Roundup Ready® Xtend® technologies. *Weed Technol* 33:569–577
- Meyer CJ, Peter F, Norsworthy JK, Beffa R (2019) Uptake, translocation, and metabolism of glyphosate, glufosinate, and dicamba mixtures in *Echinochloa crus-galli* and *Amaranthus palmeri*. *Pest Manage Sci* 76:3078–3087
- Moore KJ, Dixon PM (2015) Analysis of combined experiments revisited. *Agron J* 107:763–771
- Ou J, Thompson CR, Stahlman PW, Bloedow N, Jungulam M (2018) Reduced translocation of glyphosate and dicamba in combination contributes to poor control of *kochia scoparia*: Evidence of antagonism. *Scientific Reports* 8:5330
- Peterson MA, McMaster SA, Riechers DE, Skelton J, Stahlman PW (2016) 2,4-D past, present, and future: A review. *Weed Technol* 30:303–345
- Renton M, Willse A, Aradhya C, Tyre A, Head G (2024) Simulated herbicide mixtures delay both specialist monogenic and generalist polygenic resistance evolution in weeds. *Pest Manag Sci* 80:5983–5994
- Robinson AP, Simpson DM, Johnson WG (2012) Summer annual weed control with 2,4-D and glyphosate. *Weed Technol* 26:657–660
- Shergill LS, Barlow BR, Bish MD, Bradley KW (2018) Investigations of 2,4-D and Multiple Herbicide Resistance in a Missouri Waterhemp (*Amaranthus tuberculatus*) Population. *Weed Sci* 66: 386–394
- Shurtlee JL, Coble HD (1985) Interference of certain broadleaf weed species in soybean (*Glycine max*). *Weed Sci* 33:654–657

- Shyam C, Chahal PS, Jhala AJ, Jugulam M (2021) Management of glyphosate-resistant Palmer amaranth (*Amaranthus palmeri*) in 2,4-D-, glufosinate-, and glyphosate-resistant soybean. *Weed Technol* 35: 136–143
- Singh N, Peters TJ, Miller RP, Naeve SL, Sarangi D (2024) Profile and extent of herbicide-resistant waterhemp (*Amaranthus tuberculatus*) in Minnesota. *Weed Sci* 72: 673–682
- Staniforth DW (1965) Competitive effects of three foxtail species on soybeans. *Weeds* 13:191–193
- Steckel GJ, Wax LM, Simmons FW, Phillips WH (1997) Glufosinate efficacy on annual weeds is influenced by rate and growth stage. *Weed Technol* 11:484–488
- Stoller EW, Woolley JT (1985) Competition for light by broadleaf weeds in soybeans (*Glycine max*). *Weed Sci* 33:199–202
- Steckel GJ, Wax LM, Simmons FW, Phillips WH (1997) Glufosinate efficacy on annual weeds is influenced by rate and growth stage. *Weed Technol* 11:484–488
- Takano HK, Beffa R, Preston C, Westra P, Dayan FE (2020) A novel insight into the mechanism of action of glufosinate: how reactive oxygen species are formed. *Photosynth Res* 144:361–372

Table 1. Average temperatures and precipitation for the experiments conducted in Beresford and South Shore, SD in 2023 and 2024. Weather data is presented from the month of soybean planting until last evaluation period for each respective year.

		2023		2024	
		Average temperature (C)	Precipitation (mm)	Average temperature (C)	Precipitation (mm)
Beresford	May	18.3	41	16.7	178.6
	June	22.8	74.4	21.7	119.1
	July	21.7	128.3	22.8	20.6
	August	- ^a	-	21.1	41.4
South Shore	May	16.7	36.3	18.3	98
	June	21.7	57.9	21.1	119.1
	July	20	39.9	21.1	126
	August	-	-	19.4	55.9

^aExperiment evaluations were completed before August.

Table 2. 2,4-D (1165 g ae ha⁻¹) and glufosinate (655 g ai ha⁻¹) treatments applied to pervasive weeds in soybean at the experiment conducted at Beresford and South Shore, SD, in 2023 and 2024.

Initial	Sequential	Herbicide treatment
2,4-D	No sequential treatment	2,4-D
2,4-D	2,4-D	2,4-D fb 2,4-D
2,4-D	Glufosinate	2,4-D fb G
2,4-D	2,4-D plus glufosinate	2,4-D fb 2,4-DplusG
Glufosinate	No sequential treatment	G
Glufosinate	2,4-D	G fb 2,4-D
Glufosinate	Glufosinate	G fb G
Glufosinate	2,4-D plus glufosinate	G fb 2,4-DplusG
2,4-D plus glufosinate	No sequential treatment	2,4-DplusG
2,4-D plus glufosinate	2,4-D	2,4-DplusG fb D
2,4-D plus glufosinate	Glufosinate	2,4-DplusG fb G
2,4-D plus glufosinate	2,4-D plus glufosinate	2,4-DplusG fb 2,4-DplusG
No initial treatment	2,4-D	N fb 2,4-D
No initial treatment	Glufosinate	N fb G
No initial treatment	2,4-D plus glufosinate	N fb 2,4-DplusG

^aEach herbicide treatment was tested on common lambsquarters, common waterhemp, redroot pigweed, yellow foxtail, and velvetleaf.

^bAbbreviations: 2,4-DplusG, 2,4-D plus glufosinate; fb, followed by; G, glufosinate.

Table 3. Visual estimates of weed control with 2,4-D and glufosinate treatments 28 d after the initial herbicide application in soybean conducted in experiments at Beresford and South Shore, SD, in 2023 and 2024.

Herbicide treatment ^a	ABUTH		AMARE		AMATA		CHEAL		SETPU	
	% (SE)									
2,4-D	92 (4)	b ^{b,c}	84 (4)	e	89 (3)	cd	94 (3)	ab	- ^d	
2,4-D fb 2,4-D	99 (0.3)	a	88 (3)	de	96 (1)	ab	97 (1)	a	-	
2,4-D fb G	99 (0.125)	a	99 (0)	a	97 (1)	a	99 (0.1)	a	79 (3)	c
2,4-D fb 2,4-DplusG	99 (0)	a	99 (0.25)	a	97 (1.5)	ab	99 (0)	a	77 (5)	c
G	97 (1)	a	92 (2)	cd	66 (4)	f	82 (4)	c	58 (8)	de
G fb 2,4-D	99 (0.25)	a	99 (0.25)	ab	94 (2)	abc	97 (1)	a	63 (7)	d
G fb G	99 (0)	a	96 (2.5)	ab	98 (0.5)	a	99 (0.3)	a	92 (3)	ab
G fb 2,4-DplusG	99 (0)	a	99 (0.125)	a	99 (0.125)	a	99 (0)	a	93 (2)	ab
2,4-DplusG	99 (0.3)	a	96 (0.9)	abc	87 (4)	d	96 (3)	a	62 (9)	d
2,4-DplusG fb 2,4-D	99 (0)	a	99 (0.125)	a	96 (2)	ab	98 (1)	a	49 (3)	e
2,4-DplusG fb G	99 (0)	a	99 (0.125)	a	99 (0.4)	a	99 (0)	a	95 (2)	a
2,4-DplusG fb 2,4-DplusG	99 (0)	a	99 (0.125)	a	99 (0)	a	99 (0)	a	94 (2)	a
N fb 2,4-D	93 (2)	b	64 (5)	f	69 (3)	f	88 (5)	bc	-	
N fb G	99 (0.25)	a	94 (2)	bcd	80 (3)	e	87 (3)	c	83 (2)	bc
N fb 2,4-DplusG	98 (0.4)	a	94 (3)	abcd	92 (2)	bcd	96 (1)	a	77 (3)	c

^aAbbreviations: ‘ABUTH’, velvetleaf; ‘AMARE’, redroot pigweed; ‘AMATA’, common waterhemp; ‘CHEAL’, common lambsquarters; ‘SETPU’, yellow foxtail; fb, followed by; G, glufosinate; 2,4-DplusG, 2,4-D plus glufosinate; N, no initial treatment.

^bMeans that share the same letter within columns are not statistically different based on Fisher’s Least Significant Difference ($P < 0.05$).

^cTreatments that violated the constant variance assumption were not included in the analysis, but 95% confidence intervals were used to determine whether values were similar.

^dClethodim was applied to 2,4-D-only treatments; therefore, no data were collected for yellow foxtail.

Table 4. Weed height reduction with 2,4-D and glufosinate treatments 28 d after the initial herbicide application in soybean conducted in experiments at Beresford and South Shore, SD, in 2023 and 2024.

Herbicide treatment ^a	ABUTH		AMARE		AMATA		CHEAL		SETPU	
	% ($\pm$ SE)									
2,4-D	83 (6)	cd ^{b,c}	79 (1)	bc	68 (6)	cd	89 (4)	abc	- ^d	
2,4-D fb 2,4-D	95 (3)	abc	77 (1.5)	cd	86 (3)	ab	91 (3)	ab	-	
2,4-D fb G	98 (2)	ab	100 (0)	a	88 (4)	ab	95 (3)	a	67 (6)	bc
2,4-D fb 2,4-DplusG	100 (0)	a	96 (1.5)	a	93 (4)	a	100 (0)	a	65 (4)	bc
G	75 (10)	d	62 (2)	f	47 (5)	e	50 (1)	f	62 (5)	bc
G fb 2,4-D	93 (4)	abc	90 (2)	ab	88 (5)	ab	90 (4)	abc	71 (3)	b
G fb G	100 (0)	a	98 (0.6)	a	91 (4)	a	93 (4)	a	90 (4)	a
G fb 2,4-DplusG	99 (1)	a	96 (1)	a	94 (2)	a	97 (2)	a	91 (3)	a
2,4-DplusG	96 (3)	abc	74 (1.5)	cde	77 (6)	bc	95 (3)	a	67 (3)	bc
2,4-DplusG fb 2,4-D	96 (3)	a	99 (0.5)	a	88 (4)	ab	94 (3)	a	63 (2)	bc
2,4-DplusG fb G	97 (3)	ab	99 (0.5)	a	96 (2)	a	97 (2)	a	90 (4)	a
2,4-DplusG fb 2,4-DplusG	98 (2)	ab	99 (0.25)	a	95 (2)	a	100 (0)	a	92 (3)	a
N fb 2,4-D	72 (8)	d	74 (1)	cde	60 (3)	de	81 (5)	bcd	-	
N fb G	75 (8)	d	67 (3)	def	70 (5)	cd	80 (6)	cd	59 (3)	c
N fb 2,4-DplusG	83 (10)	bcd	65 (2)	ef	67 (4)	cd	76 (5)	e	64 (7)	bc

^aAbbreviations: ‘ABUTH’, velvetleaf; ‘AMARE’, redroot pigweed; ‘AMATA’, common waterhemp; ‘CHEAL’, common lambsquarters; ‘SETPU’, yellow foxtail; fb, followed by; G, glufosinate; 2,4-DplusG, 2,4-D plus glufosinate; N, no initial treatment.

^bMeans that share the same letter within columns are not statistically different based on Fisher’s Least Significant Difference ($P < 0.05$).

^cTreatments that violated the constant variance assumption were not included in the analysis, but 95% confidence intervals were used to determine whether values were similar.

^{dc}Clethodim was applied to 2,4-D-only treatments; therefore, no data were collected for yellow foxtail.

Table 5. Weed control and height reduction with 2,4-D and glufosinate treatments 28 d after the initial herbicide application in soybean conducted in experiments at Beresford and South Shore, SD, in 2023 and 2024.^a

Species	Herbicide treatment	Expected	Observed	P-value
<i>Control</i>		%		
ABUTH	2,4-DplusG	100	99	0.80
	N fb 2,4-DplusG	100	98	0.13
AMARE	2,4-DplusG	98	96	0.26
	N fb 2,4-DplusG	94	94	0.41
AMATA	2,4-DplusG	96	87	0.13
	N fb 2,4-DplusG	94	92	0.50
CHEAL	2,4-DplusG	99	96	0.50
	N fb 2,4-DplusG	98	96	0.30
<i>Height reduction</i>				
ABUTH	2,4-DplusG	96	96	NC
	N fb 2,4-DplusG	93	83	0.50
AMARE	2,4-DplusG	92	74	0.053
	N fb 2,4-DplusG	88	65	0.055
AMATA	2,4-DplusG	83	77	0.50
	N fb 2,4-DplusG	90	67	0.11
CHEAL	2,4-DplusG	94	95	0.80
	N fb 2,4-DplusG	96	76	0.16

^aAbbreviations: ‘ABUTH’, velvetleaf; ‘AMARE’, redroot pigweed; ‘AMATA’, common waterhemp; ‘CHEAL’, common lambsquarters; ‘SETPU’, yellow foxtail; fb, followed by; G, glufosinate; 2,4-DplusG, 2,4-D plus glufosinate; N, no initial treatment; NC, not calculated.

Table 6. Soybean yield with 2,4-D and glufosinate treatments 28 d after the initial herbicide in soybean conducted in experiments at Beresford and South Shore, SD, in 2023 and 2024.^{a,b}

Treatment	Beresford		South Shore	
	kg ha ⁻¹ (±SE)			
NTC	1064 (161)	c	766 (284)	h
2,4-D	2251 (293)	ab	2272 (280)	cdef
2,4-D fb 2,4-D	2423 (293)	ab	2130 (143)	cdef
2,4-D fb G	2431 (224)	ab	2040 (85)	efg
2,4-D fb 2,4-DplusG	2645 (228)	ab	2031 (143)	efg
G	2402 (168)	ab	2353 (195)	cde
G fb 2,4-D	2622 (221)	ab	2601 (160)	bc
G fb G	2573 (287)	ab	2918 (66)	ab
G fb 2,4-DplusG	2636 (227)	ab	3050 (77)	a
2,4-DplusG	2689 (218)	ab	2460 (192)	cde
2,4-DplusG fb 2,4-D	2844 (200)	a	2539 (154)	bcd
2,4-DplusG fb 2,4-DplusG	2497 (192)	ab	2907 (70)	ab
2,4-DplusG fb G	2302 (136)	ab	2954 (81)	ab
N fb 2,4-D	2109 (187)	b	2155 (294)	defg
N fb G	2460 (222)	ab	1890 (103)	fg
N fb 2,4-DplusG	2426 (208)	ab	1766 (88)	g

^aAbbreviations: NTC, non-treated control; fb, followed by; G, glufosinate; 2,4-DplusG, 2,4-D plus glufosinate; N, no initial treatment.

^bMeans that share the same letter within columns are not statistically different based on Fisher's Least Significant Difference ($P < 0.05$).